PCT09

RAW SEQUENCE LISTING DATE: 12/21/2001 PATENT APPLICATION: US/09/720,583A TIME: 13:24:11

Input Set : A:\MBHB00-1314 SequenceListing.txt
Output Set: N:\CRF3\12212001\I720583A.raw

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3 <110> APPLICANT: DSM N.V.
         POUWELS, Pieter
                                                                  ENTERED
 5
         van LUIJK, Nicole
         JORE, Johannes
 6
         LUITEN, Rudolf
 9 <120> TITLE OF INVENTION: Propionibacterium Vector
11 <130> FILE REFERENCE: MBHB00-1314
13 <140> CURRENT APPLICATION NUMBER: US 09/720,583A
14 <141> CURRENT FILING DATE: 2000-12-22
16 <150> PRIOR APPLICATION NUMBER: PCT/EP99/04416
17 <151> PRIOR FILING DATE: 1999-06-25
19 <150> PRIOR APPLICATION NUMBER: EP 98305033.7
20 <151> PRIOR FILING DATE: 1998-06-25
22 <160> NUMBER OF SEQ ID NOS: 13
24 <170> SOFTWARE: PatentIn version 3.0
26 <210> SEO ID NO: 1
27 <211> LENGTH: 3555
28 <212> TYPE: DNA
29 <213> ORGANISM: Propionibacterium freudenreichii LMG16545
31 <220> FEATURE:
32 <221> NAME/KEY: CDS
33 <222> LOCATION: (273)..(1184)
36 <400> SEQUENCE: 1
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39 geogeaatge eegaacaege eecagecate eettggagea ggtggeageg teaggggagt 120
41 cgggggatgt ttggcagggg atgtggaaag agagttcgct ttgctcacat ggctcaaccg 180
43 ggtaactaac tgatatgggg tettegtege ceaetttgaa caegeegagg aatggaeeac 240
45 gctgaacgtg actcgcatgc ttcactgcat gt atg gat tcg ttc gag acg ttg
46
                                       Met Asp Ser Phe Glu Thr Leu
47
49 ttc cct gag agc tgg ctg cca cgc aag ccg ctg gcg tca gcc gag aag
50 Phe Pro Glu Ser Trp Leu Pro Arg Lys Pro Leu Ala Ser Ala Glu Lys
                               15
53 tot ggg gcg tac cgg cac gtg act cgg cag agg gcg ctg gag ctg cct
                                                                      389
54 Ser Gly Ala Tyr Arg His Val Thr Arg Gln Arg Ala Leu Glu Leu Pro
55
       25
                           30
57 tac atc gaa gcg aac ccg ttg gtc atg cag tcc ttg gtc atc acc gat
                                                                      437
58 Tyr Ile Glu Ala Asn Pro Leu Val Met Gln Ser Leu Val Ile Thr Asp
                                                                      485
61 cga gat gct tcg gat gct gac tgg gcc gca gac ctc gct ggg ctg cct
62 Arg Asp Ala Ser Asp Ala Asp Trp Ala Ala Asp Leu Ala Gly Leu Pro
                   60
63
                                       65
                                                                      533
65 tea eeg tee tae gtg tee atg aac egt gte aeg aec aec gga eac ate
66 Ser Pro Ser Tyr Val Ser Met Asn Arg Val Thr Thr Gly His Ile
               75
                                   80
69 qtc tat gcc ttg aag aac cct gtg tgt ctg acc gat gcc gcg cgg cga
                                                                      581
70 Val Tyr Ala Leu Lys Asn Pro Val Cys Leu Thr Asp Ala Ala Arg Arg
```

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71			90		٠.			95					100				
73 (caa	cct	atc	aac	cta	ctc	qcc	cqc	qtc	qaq	caq	qqc	cta	tqc	qac	qtt	629
						Leu											
75		105					110	_				115		•	•		
			aac	gat	σca	tcc		aaa	cac	caa	atc	aca	aaσ	aac	сса	ctc	677
						Ser											
79		1	1			125	_ 1 _	1		3	130	_	- 4 -	_	_	135	
		acc	αcc	cat	aca	acc	ata	t.aa	aac	ccc		σac	aca	ctc	tac		725
						Thr											,
83	JC1				140		200			145					150	01 4	
	rta	cac	acc	ctc		cac	acc	ctc	gac		atc	cac	gca	cta		σασ	773
	_	_			_	His			_				_	_	_		,,,
87	Leu	лгу	ΑΙα	155	AIU	птэ	1111	Leu	160	GIU	110	1113	nia	165	110	O.Lu	
	702	aaa	220		cat	cgc	aac	ata		caa	tca	aca	atc		cac	aac	821
						Arg											021
91	на	Сту	170	FIO	ALG	Arg	ASII	175	1111	Arg	261	1111	180	СТУ	лгу	ASII	
	~+ ~	200		++~	a a	200	200		2 ± 47	+ ~ ~	~~~	+20		<i>a a a</i>	a+a	aaa	869
						acc											003
			Leu.	Pile	ASP	Thr	190	ALG	Mec	пр	нта	195	AIG	нта	vaı	AIG	
95		185	+	~~~		~~~		~~~	~ ~ ~ ~	+~~	~~~			~+ n	++-	~~~	017
						ccg											917
		ser	ттр	СТА	GIY	Pro	Val	Ald	GIU	ттр		HIS	TIII	vaı	Pile		
99 2		_ 4			4_	205					210					215	0.65
					_			_			_	_	_		_	aca	965
	HIS	116	HIS	s Let			GIU	Thi	TIE			ı AS) GIU	Pne		a Thr	
103					220					225					230		1012
					_		-									tcc	1013
	GLY	Pro	ь тег	_		ı Asn	GIU	і ьей			: ьег	ı ser	r Arg			e Ser	
107				235					240					245			1061
																cag	1061
	Arg	Trp		_	Arg	, Asn	Phe			GIU	ı Tnı	: Phe	_		Arg	g Gln	
111			250					255					260				
				_		-		_						-		ggc	1109
	Lys			e Ser	Leu	ı Arg	_		Ser	Lys	GI3	_	_	GLu	GL3	, Gly	
115		265					270					275					
																cgt	1157
		-	Gly	7 Gly	7 Ile			Gly	7 Ala	Ser	_		Ala	His	Thi	Arg	
119						285					290					295	
						, ggt				сса	cacg	rtga	acgt	ctcc	CC		1204
122	Gln	Gln	Phe	e Leu	ı Glu	ı Gly	Leu	Ser	:								
123					300												
																caccgto	
	_								-		_		_	-	_	cacge	
																gtcggg	
																ccgtaa	
																gcggtt	
																gggcat	
137	cag	caac	gga	cgtt	gaac	cc g	gtgg	gcaa	ig tg	ttac	tcag	ggg	gaca	tgc	ccag	tctgcg	1624
139	gcg	ctcg	gat	tgac	ggta	itg g	cagt	.cgtg	rc at	gcgg	cccc	acc	gtca	aac	tcat	tcaggt	1684
141	atc	agtg	aga	acco	tcat	.gg c	acco	cctc	g tg	acac	gtto	: tcg	rttgc	gat	cago	tgctgt	1744

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143 gcgtgcgggc gtgagcgttt ctacgctgcg gcgcaggaaa tcagagcttg aggctgccgg 1804
145 agggaggta gacccgtccg gttgggtggt gccactgcgt gcactcaagg tcgtttttgg 1864
147 gqtqtcaqat qaqacctcga atqcqcccqg tcatqacqct gaqttagtqg cgcaqctqcg 1924
149 ctctgagaac gagtttttac ggcgtcaggt cgagcagcag gcgcgcacga tcgaacggca 1984
151 qqctqaqqca cacqcqqtqq tctcaqcqca qctcacacqq qttqqccaqc ttqaqqccqq 2044
153 cgacgcagca gcaccgacac tggcacccgt tgaaaggccg gctccgcgac ggcggtggtg 2104
155 gcagcgtcgg tagcggtcag gatcgctctg gcgtgacgag tgtgtctggc agtgcgaaca 2164
157 gttgctcgac cagtggcagc agaagcgaga tcgctgcgtg gtgctgttcc tcggtcagtt 2224
159 cqtcqaqqac tggcgggtct tgctgcgtcc agccgatcgc ctcggcggcc aaggtcagtt 2284
161 ccaaqctqtq ccaacqcaca cqcccctcqq ctqacaqctq aqtctcqaac tqtqcaactq 2344
163 gaccggccgg aagatgcacg ttgccgaggt cgtgagtggc caagcgcacg tcaaagagtg 2404
165 ctgcttcgta gccgcgcaga aatggcagtg ctcggtcgat tcggatcggc ctgcccaggt 2464
167 acatteeggg eegettgatg aacgeeteeg egtagaageg eacegttete ggeeeggeet 2524
169 cgtgatctgt cactgtgcac gctcctctcg atggttctcg acgctaccgg agaccaccga 2584
171 cgttcatgcc cagcgcagcg acctgaaagg accaagccga gttagccgtg ctaaccgtat 2644
173 agettgetee gtegeetetg agggeaacea eetgegeage aggtgggegg eageeegege 2704
175 gcaagcgcct accgggtttg ggcacagccc ataaatcaac gcctccggtg ttgaagcgat 2764
177 cgtgtgtcac gattgctatg cttgctaccc cttcagggtt ttcgtataca caaatcaagt 2824
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181 gagcagattc aggcatgggc agacgaggcc gaggccggat acgacctgcc caaactcccc 2944
183 aagccacggc geggaegece geeegtagga gaeggteegg geaeegtegt accegtgegt 3004
185 ctcgacgcgg ccaccgttgc cgctctcaca gaacgagcaa cagccgaggg catcacgaac 3064
187 cqttcaqacq cqatccqaqc cqcaqtccac qaqtggacac gggttgcctq acctccacga 3124
189 ctcaqcacqc aagcactacc aacgagaccg gctcgacgac acggccgtgc tctacgcggc 3184
191 cacccacgtt ctcaactccc ggccactcga cgacgaagac gacccgcgcc gctggctcat 3244
193 gateggaace gacecageag geogectact egaactegte geactgatet acgaegaegg 3304
195 ctacqaactq atcatccacq caatqaaaqc ccqcacccaa tacctcqacc agctctaacc 3364
197 aagaaaggaa cctgatgagc gaccagctag acagcgaccg caactacgac ccgatgatct 3424
199 tegacgtgat gegegagaee gegaacegeg tegtegeeae gtaegttgea tgggaagatg 3484
201 aageegetga teeeegegag getgegeaet ggeaggeega gegatteege aeeeggeaeg 3544
203 aggtgcgcgc c
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206 <210> SEQ ID NO: 2
207 <211> LENGTH: 303
208 <212> TYPE: PRT
209 <213> ORGANISM: Propionibacterium freudenreichii LMG16545
211 <400> SEQUENCE: 2
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215 Pro Leu Ala Ser Ala Glu Lys Ser Gly Ala Tyr Arg His Val Thr Arg
216
                                    25
218 Gln Arg Ala Leu Glu Leu Pro Tyr Ile Glu Ala Asn Pro Leu Val Met
219
                                40
221 Gln Ser Leu Val Ile Thr Asp Arg Asp Ala Ser Asp Ala Asp Trp Ala
222
                            55
                                                60
224 Ala Asp Leu Ala Gly Leu Pro Ser Pro Ser Tyr Val Ser Met Asn Arg
                        70
                                            75
227 Val Thr Thr Thr Gly His Ile Val Tyr Ala Leu Lys Asn Pro Val Cys
228
                                        90
230 Leu Thr Asp Ala Ala Arg Arg Pro Ile Asn Leu Leu Ala Arg Val
```

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100
                                     105
231
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233 Glu Gln Gly Leu Cys Asp Val Leu Gly Gly Asp Ala Ser Tyr Gly His
                                120
            115
236 Arg Ile Thr Lys Asn Pro Leu Ser Thr Ala His Ala Thr Leu Trp Gly
                            135
                                                 140
239 Pro Ala Asp Ala Leu Tyr Glu Leu Arg Ala Leu Ala His Thr Leu Asp
                        150
                                             155
240 145
242 Glu Ile His Ala Leu Pro Glu Ala Gly Asn Pro Arg Arg Asn Val Thr
                                         170
                    165 .
245 Arg Ser Thr Val Gly Arg Asn Val Thr Leu Phe Asp Thr Thr Arg Met
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                                     185
248 Trp Ala Tyr Arg Ala Val Arg His Ser Trp Gly Gly Pro Val Ala Glu
                                 200
            195
251 Trp Glu His Thr Val Phe Glu His Ile His Leu Leu Asn Glu Thr Ile
        210
                            215
254 Ile Ala Asp Glu Phe Ala Thr Gly Pro Leu Gly Leu Asn Glu Leu Lys
                                             235
257 His Leu Ser Arg Ser Ile Ser Arg Trp Val Trp Arg Asn Phe Thr Pro
                    245
                                         250
260 Glu Thr Phe Arg Ala Arg Gln Lys Ala Ile Ser Leu Arg Gly Ala Ser
                260
                                     265
263 Lys Gly Gly Lys Glu Gly Gly His Lys Gly Gly Ile Ala Ser Gly Ala
            275
                                280
266 Ser Arg Arg Ala His Thr Arg Gln Gln Phe Leu Glu Gly Leu Ser
        290
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270 <210> SEQ ID NO: 3
271 <211> LENGTH: 85
272 <212> TYPE: PRT
273 <213> ORGANISM: Propionibacterium freudenreichii LMG16545
275 <400> SEQUENCE: 3
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279 Ala Ala Lys Lys Leu Gly Val Ser Glu Ser Thr Val Lys Arg Trp Thr
                                      25
282 Ser Glu Pro Arg Glu Glu Phe Val Ala Arg Val Ala Ala Arg His Ala
                                 40
285 Arg Ile Arg Glu Leu Arg Ser Glu Gly Gln Ser Met Arg Ala Ile Ala
                             55
288 Ala Glu Val Gly Val Ser Val Gly Thr Val His Tyr Ala Leu Asn Lys
                         70
                                              75
289 65
291 Asn Arg Thr Asp Ala
295 <210> SEQ ID NO: 4
296 <211> LENGTH: 59
297 <212> TYPE: DNA
298 <213> ORGANISM: Artificial Sequence
300 <220> FEATURE:
301 <223> OTHER INFORMATION: Synthetic Duplex DNA designed to link EcoRI and AvaI
303 <400> SEQUENCE: 4
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Input Set : A:\MBHB00-1314 SequenceListing.txt
Output Set: N:\CRF3\12212001\I720583A.raw

304 aattcaaget tgtcgacgtt aacctgcagg catgcggatc cggtaccgat atcagatct 59 307 <210> SEQ ID NO: 5 308 <211> LENGTH: 59 309 <212> TYPE: DNA 310 <213> ORGANISM: Artificial Sequence 312 <220> FEATURE: 313 <223> OTHER INFORMATION: Synthetic Duplex DNA designed to link EcoRI and AvaI 315 <400> SEQUENCE: 5 316 ccgaagatct gatatcggta ccggatccgc atgcctgcag gttaacgtcg acaagcttg 59 319 <210> SEQ ID NO: 6 320 <211> LENGTH: 24 321 <212> TYPE: DNA 322 <213> ORGANISM: Artificial Sequence 324 <220> FEATURE: 325 <223> OTHER INFORMATION: Synthetic DNA designed to create Acc65I and BglII ends 327 <400> SEQUENCE: 6 24 328 gtaccggccg ctgcggccaa gctt 331 <210> SEQ ID NO: 7 332 <211> LENGTH: 24 333 <212> TYPE: DNA 334 <213> ORGANISM: Artificial Sequence 336 <220> FEATURE: 337 <223> OTHER INFORMATION: Synthetic DNA designed to create Acc65I and BglII ends 339 <400> SEQUENCE: 7 24 340 gatcaagctt ggccgcagcg gccg 343 <210> SEQ ID NO: 8 344 <211> LENGTH: 35 345 <212> TYPE: DNA 346 <213> ORGANISM: Artificial Sequence 348 <220> FEATURE: 349 <223> OTHER INFORMATION: Synthetic primer for erythromycin resistance 351 <400> SEQUENCE: 8 35, 352 aaactgcage tgctggcttg cgcccgatgc tagtc 355 <210> SEQ ID NO: 9 356 <211> LENGTH: 76 357 <212> TYPE: DNA 358 <213> ORGANISM: Artificial Sequence 360 <220> FEATURE: 361 <223> OTHER INFORMATION: Synthetic primer for erythromycin resistance 363 <400> SEQUENCE: 9 364 aaactgcagc agctgggcag gccgctggac ggcctgccct cgagctcgtc tagaatgtgc 60 366 tgccgatcct ggttgc 369 <210> SEQ ID NO: 10 370 <211> LENGTH: 32 371 <212> TYPE: DNA 372 <213> ORGANISM: Artificial Sequence 374 <220> FEATURE:

377 <400> SEQUENCE: 10

375 <223> OTHER INFORMATION: Synthetic primer for cobA

VERIFICATION SUMMARY

DATE: 12/21/2001

PATENT APPLICATION: US/09/720,583A

TIME: 13:24:12

Input Set : A:\MBHB00-1314 SequenceListing.txt
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